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(54) Processing particulate material

(57) Different particle types are removed from a liquid as it flows through a plurality of chambers (103, 115) connected in series. In each chamber there is maintained an ultrasonic standing wave which moves perpendicularly to the wave fronts and carries particles towards certain regions, different frequencies in the respective chambers attracting different particles. In Fig. 1 diluted blood is treated in chamber 103, where red copuscles are pushed to the right-hand end, and the liquid then passes to chamber 115 where bacteria are urged against impervious window 120, where they may be observed through magnifying system 124. The process may be operated continuously or batch-wise. In Fig. 2 (not shown) the waves are propagated at right angles to the liquid flow, upwards in chambers where a thermal gradient prevents the liquid flow from disturbing the separated particles.

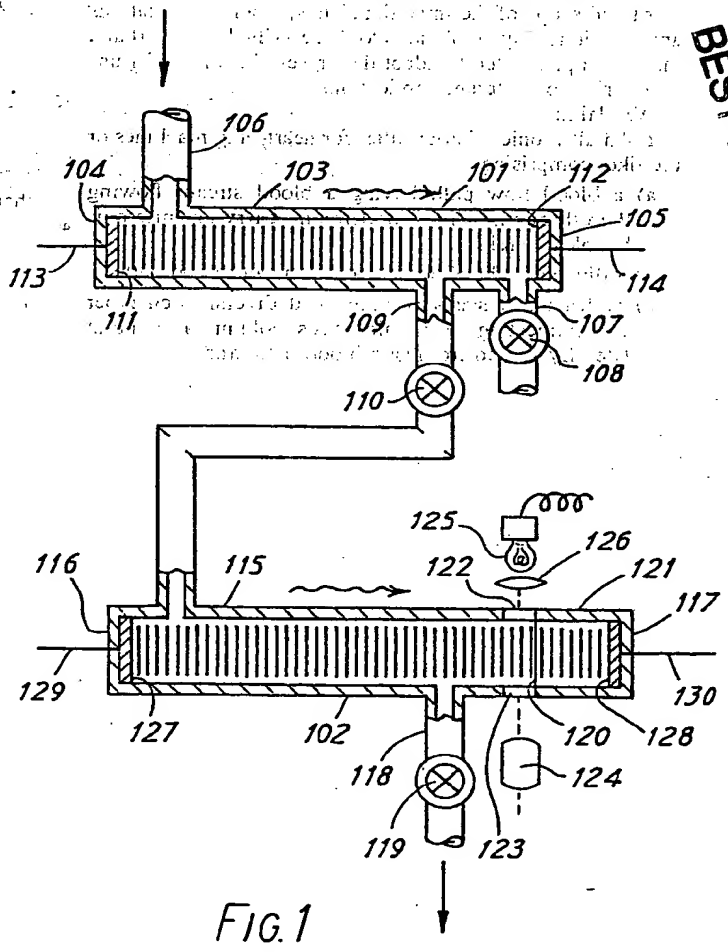

FIG. 1

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The drawings originally filed were informal and the print here reproduced is taken from a later filed formal copy.

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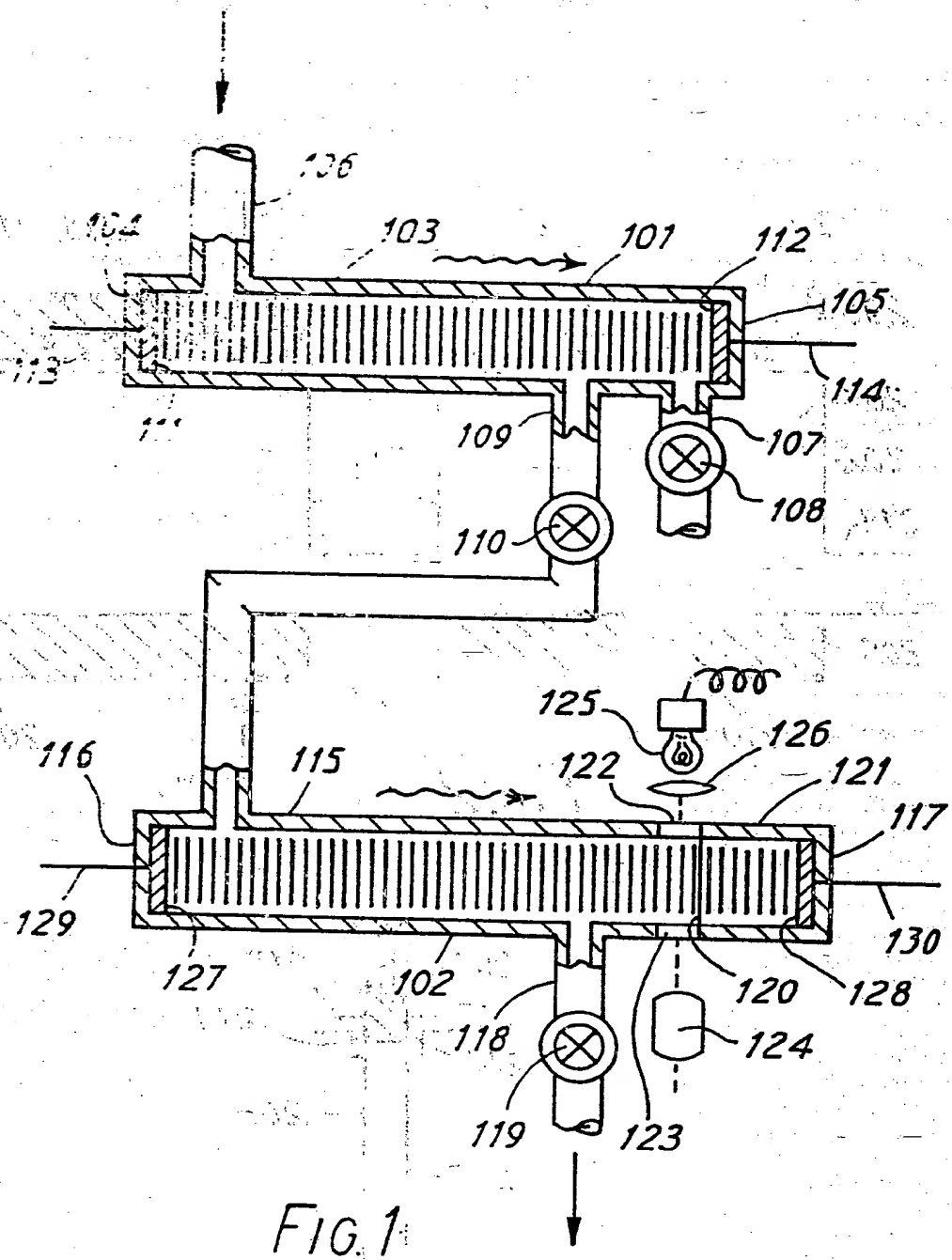

FIG. 1

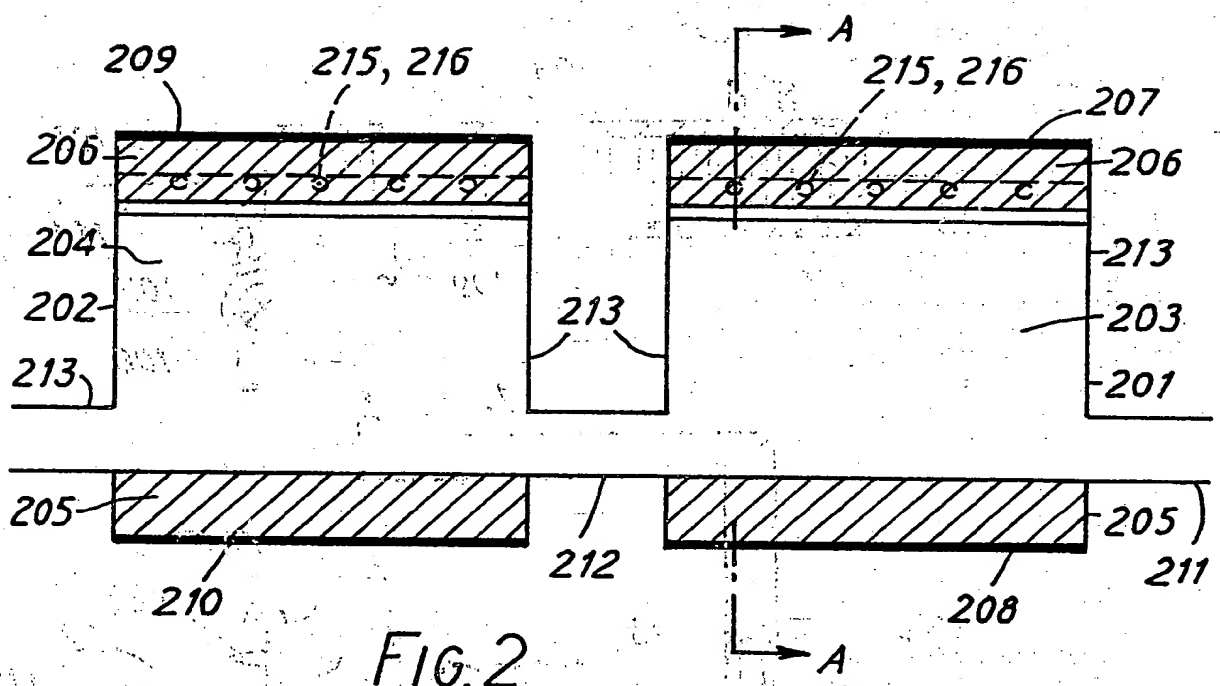

FIG. 2

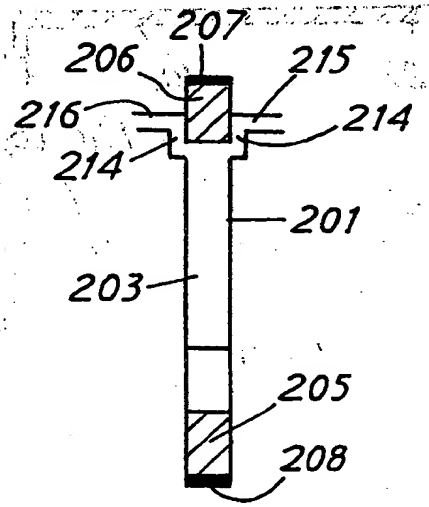

FIG. 3

SPECIFICATION

Processing Particulat Material

The present invention relates to the manipulation of particulate matter in a fluid medium. In a particular field of application, it relates to the processing of body fluids, for example for diagnostic analysis or routine health care purposes, but it can be utilised in other fields.

The successful diagnosis of many diseases often requires the confident detection of small populations of micro-organisms present in the body fluids of the patient. For example, such detection may require finding and identifying quite small populations of bacteria, such as 10^4 cells per ml of *E. coli* present in pathological urine also containing much other particulate matter such as cell debris. A further example is the problem of detecting extremely small numbers of micro-organisms in blood. The only practical method presently available for achieving this is that of culturing the blood. Minute populations of pathological micro-organisms in blood may be highly significant in diagnosis, but attempts to detect these small numbers of bacteria by culture techniques are frequently ineffective and often are very prolonged, and this at a time when the information is needed in the ward in hours, rather than days.

In typical current medical practice, five blood cultures are set up at intervals during a period of 2 days and all samples are cultured both aerobically and anaerobically for many days. Many blood pathogens are fastidious bacteria which can be grown only under special conditions and then only with difficulty. Delay and high cost are inevitable in procuring this vital diagnostic information via this route. There is a clear need for a technique that will enable minute populations of micro-organisms present in body fluids, to be concentrated and, detected rapidly and easily from relatively small quantities of such body fluids.

The use of a standing wave generated by an ultrasonic frequency emission for gas column chromatography of blood samples is known from G.B. 2 059 796A. The technique proposed there has a number of serious limitations, however. One particular difficulty is that it requires the injection of a particulate sample into a gas stream and the control of the separation rate in the carrier gas flow along a chromatographic column while maintaining the particles suspended therein, so as to introduce a spatial separation between different particle types.

According to one aspect of the present invention, there is provided a method of separating a plurality of particle types present in a fluid in which the fluid is passed through a plurality of chambers in series, in each chamber there being an ultrasonic standing wave that is moved transversely to its wave fronts, whereby said wave fronts are progressively displaced towards a chosen end region of the chamber, the conditions in the respective chambers being controlled to cause a selected type of particle in the fluid to become attached to the standing wave and be displaced towards said end region of each chamber and be concentrated at and/or collected from said end region, the successive chambers thereby separating the different selected particle types in the fluid.

Although we have referred so far to the problem of detecting micro-organisms it will be apparent that the invention has a more general applicability. Thus it may be employed as a chromatographic technique for separating and/or concentrating distinct particle types from a fluid, not only a body fluid such as blood, which contain a diverse population of particulate matter.

In any of the chambers, the particles may be concentrated from a static body of fluid filling the chamber or from a fluid flowing through the chamber. The concentration of particles at an end region of a chamber may be extracted from the chamber for examination or use, or inspection and/or analysis of the particles can be performed while they are retained at the end region. If desired, convergent standing waves, and/or a plurality of non-coaxial, intersecting standing waves, can be arranged to focus the retained particulate matter into a chosen location, thus amplifying the concentrating ability of the technique and enhancing detection/identification of the particulate matter. Even if a particular particle type is present only in small quantities, after a sufficient volume of fluid has passed through the zone in which the standing wave is established it will be found that the concentration of the selected particles will be sufficient to enable determination and identification to be performed. This can be effected by any appropriate conventional technique and does not form part of the invention.

According to a further aspect of the invention, there is provided an apparatus for separating and a plurality of particle types present in a fluid flow, comprising a plurality of chambers arranged to receive a quantity of fluid in sequence, means for establishing in each chamber an ultrasonic standing wave and for moving said standing wave transversely to its wave fronts, whereby said wave fronts can be progressively displaced towards a chosen end region of the chamber, means for controlling the conditions in the respective chambers to cause selected particle types in the fluid to become attached to the standing wave so that the different selected particle types in the fluid can be concentrated at and/or collected from said end regions of the respective chambers.

A standing wave pattern results, as is well known, from the superimposition of two progressive waves outputs, the standing wave occurring in the area of overlap of the two progressive waves whether or not the axes of propagation of the outputs are coincident. If the two opposing progressive waves differ in amplitude the net effect is a standing wave upon which is superimposed a progressive element. In practice this will almost always arise to some degree due to the attenuation of ultrasound in the fluid medium. If the two progressive waves differ slightly in frequency, the standing wave will move towards the source emitting the lower frequency. If the two opposing waves differ both in frequency and in amplitude a combination of these effects will

be observed, and by control of the sources each effect can be regulated to achieve th desired result.

A drifting standing wave can therefore be established by having two opposing sources of ultrasound (i.e. transducers) operating at fixed frequencies differing slightly one from the other. In practice, such an arrangement can be difficult to maintain unless all possible coupling between the two sources is eliminated or overridden. If coupling remains possible, there will be a tendency for the two sources to "lock" onto the same frequency. However, a controllable drift of the standing wave can be established by progressively altering a phase difference between the signals fed to the two opposing sources of ultrasound.

A primary requirement of the invention is that the standing wave established within the fluid medium should exert on one or more types of particles within the fluid medium a discernable tendency to collect at the nodes of the standing wave. In this respect there will be a relationship between the physical properties of the particles involved and the wavelength and energy of the standing wave. A major physical property of the particles that will determine this relationship is their maximum dimension, but many other factors come into consideration however, as for example density and shape.

As a means for separating dissimilar particles present in the fluid medium, the invention utilises a phenomenon which we shall term herein as "nodal delay". A particle suspended in the fluid and located in a moving standing wave will move with the nodes of the standing wave if the acoustic forces predominate, whereas if subject to Stokes forces due to movement of the liquid and these viscous, drag forces predominate it will move with a mean velocity equal to the liquid velocity. If the peak acoustic forces are almost exactly balanced by the non-acoustic forces, the particle will move at times with the same velocity as the nodes, and at other times will move with a complex oscillating motion in which, however, the standing wave does not impart any net motion to the particle. In practice the acoustic energy density of each part of a nodal array will not be exactly equal, nor may it be possible to have non-acoustic forces with a constant magnitude in every part of a system. For such reasons, when the opposing forces are nearly in balance, a given particle will at one moment move with the standing wave but at another moment will not move with the wave.

The "nodal delay" is the proportion of the time during which the given particle is moving with the wave. Thus, if the tendency of a given particle type to mass at the nodes overrides totally the effect of the opposing forces in the fluid medium, the nodal delay then experi nced by that particle type is unity. If the tendency of a given particle type to remain at the nodes is overriden totally by the effect of the opposing forces, the nodal delay experienced by that particle type can be said to be zero. Nodal delays having intermediate values arise only when the acoustic and the non-acoustic forces are nearly in balance.

In this explanation, those stations along the axis of propagation of the acoustic en rgy which the adjacent net acoustic forces act towards are termed nodes and those which said acoustic forces act away from are termed antinodes, and it is indicated that the particles influenced by a standing wave will tend to accumulate at the nodes of the wave. It must be said, however, that the detailed theory underlying the observed phenomenon of standing waves and their effect of particles in fact is not fully understood, but any lack of theoretical understanding has no bearing on the practical application of the invention. In particular, unless stated otherwise, the term "nodes" is used herein may thus be considered to include both nodes and antinodes, because in practice it does not appear to matter to the results that can be achieved.

Considering in more detail the acoustic forces acting on a particle that is small compared to the wavelength of a stationary standing wave having no progressive element, if these forces predominate they urge the particle to the nearest node. Thus in the absence of any applied opposing force, the particle moves to the node at a velocity proportional to the magnitude of the net acoustic force on it at any instant—i.e. the particle has acoustic forces acting on it which are at a maximum when the particle is at a point halfway between a node and an adjacent antinode.

Suppose now that there is an opposing force from the relative movement of a liquid axially to the standing wave at a constant velocity, the force being less than the maximum acoustic force at that intermediate point. The equilibrium position of the particle is no longer coincident with the node but is displaced somewhat downstream, although it still remains attached to the force field of the node. If the opposing force becomes larger (the velocity increases), so that it exceeds the maximum acoustic force, the particle is swept through the standing wave, accelerating as it nears a node and slowing after it has passed the node, with the result that the particle mean velocity is the same as if no standing wave were present. Thus, a particle is either held on a node or it travels at a mean velocity which is substantially identical with the velocity of the fluid.

If two different particles are considered having different acoustic responses, under the influence of the opposing non-acoustic force they will move to different positions of displacement from a node while they are held by standing wave. As the opposing force is increased, so one particle will be released while the other remains held, so that a relative axial movement can be imposed betwe n particles of two different types.

Many alternative forms of non-acoustic force can be used. For example, th effect of the standing wave can be opposed by gravity or an enhanced gravitational field. In the case of gravity, separation is not dependent on size but on the other acoustic properties of the particles. As further examples other field forces such as electrostatic or electromagn tic forces may be appropriate.

Using an axial opposing force such as th viscous drag of a liquid, in one extreme condition the

position of the standing wave is fixed (i.e. the array of nodes forming the standing wave is at least substantially motionless relative to the sources generating the standing wave) and the carrier liquid flows through the standing wave. In this condition the nodes of the standing wave can be likened to a series of filters or grids holding particles of a selected type through which the liquid passes with any particles that are less strongly influenced by the acoustic forces. At the alternative extreme, with the liquid stationary and the standing wave caused to drift along its axis, the nodes of the standing wave can be likened to a comb that is passed through the fluid medium carrying with it those particles which are held at the nodes. It will be understood that by combination of the movements of both the fluid and the standing wave, as well as by controlling the standing wave intensity and/or frequency, it is readily possible to achieve an extremely wide degree of control. Between these extremes are systems in which both the fluid and standing wave move.

From the above discussion, it might be implied that the non-acoustic forces act in a direction parallel to the axis of a standing wave but this is not the only form of force field which may be concerned. The present invention also comprises arrangements in which there is a component of fluid flow normal to the axis of the standing wave, in an extreme case the fluid flow being substantially at right angles to the standing wave axis. It is therefore necessary to consider also the effect of forces acting normal to the axis of the standing wave.

A particle influenced by the standing wave will not only move towards a node but because the acoustic energy density in the nodal plane will generally not be uniform, it will move parallel to the plane in the direction of increasing energy density. The energy density gradient in the plane of the node is very much smaller than the gradient axially of the standing wave, especially at the ultrasonic frequencies used in the present invention. It can thus be displaced parallel to the nodal plane by a much smaller non-acoustic force, so it is easier to detach the particle transversely from a standing wave by a liquid flow normal to the axis of the wave than to detach it axially. The work done to remove the particle parallel to the nodal plane is the same as moving it axially from a node halfway to an adjacent antinode however, because the displacement required parallel to the nodal plane is much greater than the displacement transverse thereto.

Under practical conditions, the velocity with which a particle moves in the plane of a node is not always the same as the liquid flow velocity in this direction; the particle will accelerate as it moves up an acoustic energy density gradient and will slow in a decreasing gradient. As described with axial movement, however, the average velocity is that of the velocity of the carrier fluid in the plane of the node, unless the particle is held by the node at some local high acoustic energy density region from which the Stokes forces cannot dislodge it for a time. Finite nodal delays therefore can operate in directions both along and transverse to the axis of the standing wave but in general have no practical effect if a two part separation is being performed within an enclosed chamber.

In the analysis of blood using the process of the present invention, by timing the frequency and/or amplitude of the standing wave it is possible to retain, for example, a relevant group of blood particulates without significantly impeding the passage of micro-organisms, or, at higher frequencies, to retain a relevant micro-organism, without significantly impeding the passage of other particulate material in the fluid. Therefore, a preliminary separation, e.g. of red blood cells, in a first chamber can facilitate the subsequent concentration of the micro-organism from the residual body fluid. The preliminary separation can be effected using a standing wave according to the invention, tuned to a frequency and amplitude appropriate to entrap the larger particulate matter in that chamber but to permit easier passage of the micro-organism and the smaller body fluid particles for separation of the micro-organism in a succeeding chamber.

The invention can be applied to any body fluid, such as blood, lymph, cerebro-spinal fluid, amniotic fluid and urine. The particulate matter that can be selectively concentrated by means of the invention can be any such matter encountered in body fluids, e.g. red blood corpuscles, white corpuscles, platelets, protozoa, bacteria and viruses, as well as other biological particles. More generally, it has a still wider application embracing both organic and non-organic particulate material in all kinds of fluid media.

The upper size limit for manipulation of particles by the method of the invention is determined by the internodal distance in the standing wave. Acoustic forces on the particles essentially cease to be effective when the particle is so large that it spans the distance between a node and the adjacent antinode. In water at NTP, with directly opposed ultrasonic sources at 100kHz, this distance is about 3.5mm.

The lower limit to size is more difficult to specify because of the number of factors involved and requires to be determined experimentally in any particular case. It appears that one factor is the size of particles relative to the internodal distance, because the efficiency of separation falls as the difference between these dimensions increases.

For the types of particles exemplified above, the optimum frequency needed to establish an appropriate standing wave in water will be in the range 100kHz to as high as 250MHz, or more usually 100MHz. In the upper end of this range it is necessary to consider the reduction of intensity of the ultrasonic radiation with distance from the source: such attenuation means that the two progressive waves cannot be identical in amplitude except in a very small region and in practice the standing wave will include a minor progressive component. The use of liquid, e.g. water, as a carrier medium enables such frequencies to be employed without undue attenuation and a useful operating zone extends to both sides of the position of energy

balance; for example, at 1MHz the intensity of a propagated wave is halved only after travelling 13.8 metres and at 5MHz the corresponding figure is 0.55 metres. For many practical purposes it is only necessary to establish the standing wave over much smaller distances. Using the example of directly opposed sources operating in water, at 100MHz, the distance between a node and an adjacent antinode is about 4 microns, but this should still allow large macromolecules with molecular weight in the region of 10^6 Daltons (equivalent to a sphere of about 0.005 microns diameter) to be separated. The working distance will of course be small due to the attenuation at that frequency, but an axial length of 1mm (133 nodes) may be available.

If a carrier liquid is to be added to the material being treated, the use of water has an advantage in that it is a benign medium for biological particles generally. In addition, in some instances it is an advantage to be able to choose a medium with a density not widely different from those of particulate materials required to be separated, so that gravitational effects can be minimised and sensitivity to the differences between particle types is improved. At higher frequencies there are also the advantages of a reduction of beam spread and the ability to use higher powers without risk of cavitation. It has been found, moreover, that the efficiency of association of particles with nodes increases with frequency.

While in most cases water is the most appropriate medium it may be advantageous to increase the liquid density by the addition of solutes such as sucrose, or to control osmotic effects, or to increase viscosity by the addition of suitable macromolecular materials. Polar liquids having a lower density than water, such as methyl alcohol, may be useful carrier liquids for some particulate materials, while for the analysis of substances sensitive to polar liquids, such as cereal flours, or for water-sensitive powders such as cements, non-polar liquids such as kerosene would be chosen. In other cases it may be advantageous to use oils having a higher viscosity and lower density than water. It will be understood that the different characteristics of such diverse liquids will be a factor in determining the frequencies of the ultrasonic sources employed.

It should also be understood that the invention may be embodied in the applications in which the carrier liquid has a higher density than the particulate material. Thus, a high density mobile liquid such as bromoform may be suitable to some particular cases. As further possibilities, it will be appreciated that the method of the present invention can be operated at temperatures both considerably higher and lower than room t mperatur , and the static pr ssure may also be controlled, so that substances normally solid or gaseous at normal temperatures and pressures can also be considered as carrier liquids. In the case of inorganic particles, it may be preferred to use liquid carriers other than water, e.g. for density considerations or in order to ensure that the particulate material is dispersed.

By way of example, an apparatus according to the invention will be described with reference to th accompanying schematic drawings in which:

Fig. 1 illustrates in section an apparatus according to the invention,

Figs. 2 illustrates in section a further apparatus according to the invention, and

Fig. 3 is a sectional view in the plane A—A in Fig. 1.

The apparatus of Fig. 1, intended for blood analysis, comprises two particle-trapping units 101 and 102 connected in series. The first unit 101 comprises a chamber in the form of a horizontal cylindrical column 103 closed at both ends 104, 105. An inlet 106 leads into column 103 adjacent the end 104 and an outlet 107 closable by means of valve 108 leads from column 103 adjacent to the opposite end 105. Approximately two-thirds of the way along column 103 from end 104 a further outlet 109 closable by means of valve 110 leads to the second particle-trapping unit 102. A matched pair of ultrasonic transducers 111 and 112 are mounted at the ends 104 and 105 respectively of column 103 such that a standing wave can be generated along the cylindrical axis of the column, the transducers 111 and 112 being connected via transmitting means 113 and 114 respectively, to ultrasonic generators (not shown). The nodes of the standing wave established within the column 103 will extend perpendicular to the axis of the column. Transducers 111 and 112 extend over the entire bor of the column 103. Fluid flowing through the column from inlet 106 to outlet 109 thus passes through the standing wave.

The second particle-trapping unit 102 comprises a similar long horizontal cylindrical chamber or column 115 closed at the both ends 116 and 117. Outlet conduit 109 from the first column 103 enters column 115 adjacent to the end 116. An outlet 118, closable by means of valve 119, emerges from the column 115 at a position approximately two-thirds of the way along from the end 116. An ultrasonic window 120 is mounted within the column 115 between the outlet 118 and the end 117, completely closing the bore of the column 115 and sealing off a short length 121 of the column adjacent the extreme end 117. This sealed portion 121 of the column 115 contains water, or a material transparent to ultrasound and having the same acoustic properties as water. The wall of the unsealed portion of the column 115 immediately adjacent to the ultrasonic window 120 is pierced by a pair of observation windows 122 and 123 made from glass or similar light-transmitting material. Observation windows 122 and 123 provide means for enabling particulate material accumulated against the ultrasonic window to be observed through a magnifying optical system 124, when suitably illuminated. The illumination system illustrated comprises a simple light source 125 and a focusing lens 126, but alternative illumination means can be used.

A matched pair of ultrasonic transducers 127 and 128 are mounted at the ends 116 und 117 r spectively of the vessel 115. The transducers are connected via transmitting means 129 and 130

respectively to ultrasonic signal generators (not shown) and are mounted such that an ultras nic standing wave can be established between them extending along the entire cylindrical axis of the column extending through the ultrasonic window 120. Each transducer 127 and 128 extends over the entire transverse area of the column and hence fluid flowing through the column from inlet conduit 109 to outlet 118 passes through the standing wave.

As depicted, the apparatus can be operated in either a batchwise or continuous manner. A blood sample, diluted as necessary with a suitable carrier fluid such as phosphate buffered saline, is admitted to the column 103 via inlet 106. A continuous flow through the entire apparatus can be maintained by means of a pump (not shown), or a sample of blood can be fed into the column 103 after which valves 108 and 110 would be closed thus maintaining the sample in a static situation within the column 103. Transducers 111 and 112 are activated and a standing wave established between them. The frequency and/or amplitude of this standing wave should be such that larger particulate matter within the blood sample, e.g. red blood corpuscles, will be restrained at the nodes of the standing wave but smaller particles, such as micro-organisms if present in the blood sample, will remain unaffected by the standing wave. By progressive changes of its phase the effective frequency of the signal fed to transducer 111 is slightly higher than that fed to transducer 112 the nodes of the standing wave can be made to move slowly towards the end 105 of the column 103. This movement will progressively carry all of the entrained larger blood particles towards the right hand end 105 and thus beyond outlet pipe 109. Within a short space of time all of the larger particles in the blood sample will be constrained at the extreme right hand end of the column 103 adjacent outlet pipe 107. The bulk of the blood sample within the column will therefore have been cleared of these larger particles, and will comprise simply fluid material and smaller particulate matter in the sample. This blood fraction devoid of larger particulate matter can be transferred via conduit 109 into the column 115 for further analysis. Preferably, this transmission between columns 103 and 105 is conducted while the moving standing wave in column 103 is maintained thus avoiding contamination with the larger particulate matter as the cleared fraction is drawn from the column 103. The accumulated larger particulate matter can thereafter be drained from the column 103 by opening valve 108. If the process is being operated in a continuous manner the accumulated matter can be flushed out by a flow through a further inlet (not shown) opposite the outlet 107.

When the clear d blood fraction has been transferred to the column 115, transducers 127 and 128 can be activated and an ultrasonic standing wave established between them. The frequency and/or amplitude of this second standing wave should b such that the smaller particulate matter remaining in the blood sampl will be held at the nodes of the standing wav . At this stage of th process, the tuning of the standing wave can be sufficiently critical that only one specific type of small particulate matter, e.g. a particular species or type of micro-organism, will be significantly affected by the standing wave. By arranging that the effective frequency of the signal fed to transducer 127 is marginally higher than that of the signal fed to transducer 128, the standing wave can be made to move slowly towards the end 117 of the column 115. This movement will carry the entrained particulat matter with it towards that end of the column. The moving wave will pass through ultrasonic window 120, but the entrained particulate matter will accumulate against the window as long as the standing wave is maintained. Within a short space of time all of the particulate matter that is significantly affected by the second standing wave will have been cleared from the bulk of the residual blood sample and will have collected in the immediate vicinity of the ultrasonic window 120. The presence of this accumulated particulate matter can be detected for example by suitable visible or ultraviolet illumination from light source 125.

By using this method, minute populations of small particulate materials, such as micro-organisms in a blood sample, can be detected. The accumulation of the particulate matter in the vicinity of the ultrasonic window will provide a local concentration so considerably facilitating detection. Larger particulate matter in the original blood sample that might otherwise have masked the presence of the smaller particulate matter, will already have been removed from the sample in th earlier separation procedure conducted in column 103. The method provides a more rapid analysis of blood or other body fluids and one which is much less costly in terms of energy and equipment than conventional techniques relying, for example, on the use of centrifuges.

In the further example of Fig. 2, the apparatus again comprises two particle trapping units 201, 202 connected in series. These units are of identical construction, and each comprises a rectangular chamber 203, 204 of identical form, with a base 205 and a top wall 206 able to transmit ultrasound from respective pairs of opposed barium titanate transducers 207, 208 and 209, 210 attached to them by means providing an efficient coupling for the ultrasound. Preferably the base and top wall of each chamber are also efficient heat conductors; they may be made, for example, from aluminium. Th first chamber has large cross-section inlet and outlet ports 211, 212 disposed in opposite end walls 213 immediately adjacent the base 205. The port 212 also serves as an inlet port to the second chamber which also has an outlet port 213. At the top of each chamber, manifolds 214 (Fig. 3) extending slightly below the level of the top wall have rows of small inlet and outlet ports 215, 216 opening into th m, whereby a fluid flow can be directed across the entire width of the top wall 206. The porting 215, 216 thus allows collected particulat matter to be removed fr m each chamber by a flushing flow across the top wall 206 from one set of ports to the other. It is, of course, possible to arrange f r examination of the material collected in the top of

each chamber in situ, as is also done in the first example.

In operation, the pair of transducers of each unit are powered to produce a standing wave in each chamber with nodes extending parallel to the base and top wall 205, 206 and moving upwardly towards the top wall at a slow rate, across a flow of liquid through the chamber from inlet 211 or 212 to outlet 212 or 213. Particles that become attached to the nodes of the standing wave are thus carried upwards to the top wall where they can be inspected while held in the chamber or be collected from the chamber. As in the preceding example, the frequency and amplitude of the standing wave will be so adjusted for each chamber that different selected particle types can be collected from the fluid in the respective chambers. In the blood analysis process described in that first example, therefore, the red blood corpuscles and other larger particles will be concentrated in the chamber 203 while the micro-organism sought will be concentrated in the chamber 204 if present.

In order to ensure that the main liquid flow introduced at the bottom of each chamber does not diffuse upwards and so carry particles not influenced by the acoustic energy up towards the top wall, it may be required to adopt measures that prevent such diffusion of the liquid into the upper part of the chamber. This can be achieved by establishing a small density gradient in which carrier liquid entering through the inlet to the chamber is denser than the liquid in the upper region of the chamber, so that a stratified liquid flow is obtained.

Thus, means may be provided to cool the bottom wall of a chamber and warm the top wall whereby liquid within the chamber is cooled from below and warmed from above to generate a density gradient. The flushing flow between the ports 215, 216 is provided by a warmed liquid and the existence of the density gradient ensures that it will not flow downwards to disturb the main liquid flow in the lower region of the chamber. The density gradient also assists in controlling the flow of liquid between the main inlet and outlet ports, this flow being kept sufficiently cool to prevent it drifting into the warmed and therefore less dense liquid contained in the chamber above the level of the ports.

It is also possible to operate the apparatus isothermally and retain the right density gradient by employing a liquid in the main body of the chamber and for the flushing flow that is slightly less dense than the carrier liquid from which the particles are to be separated. For example, if the carrier liquid is water, above it and for the flushing flow there can be used water which has had a small addition of ethyl alcohol.

It will be appreciated that flushing of collected particles can be performed continuously or at intervals. The main liquid flow may be maintained for continuous operation of the separation process, but alternatively batch processing can be performed with the main liquid flow continuous or discontinuous, and indeed the fluid can be simply held in a chamber while the separation process is carried out. Although both examples show two-chamber apparatus, it will be understood that further chambers can be connected in series if a larger number of particle types are to be collected, and also that mixed combinations can be used with one or more chambers having the standing wave and fluid flow substantially coaxial, as in Fig. 1 and one or more other chambers having the flow transversely to the standing wave axis, as in Fig. 2.

The present invention also includes method of and apparatus for separation in a single chamber, such as one of the chambers illustrated in Figs. 2 and 3, in which a fluid flow is directed across an end region of the chamber and is substantially confined to said region, while separated particles are influenced by the standing wave to be moved away from said region to an opposite end region of the chamber, whether for concentration there or for removal by a cross flow in a direction transverse to said fluid flow in the direction of movement of the standing wave.

CLAIMS

1. A method of separating a plurality of particle types present in a fluid in which the fluid is passed through a plurality of chambers in series, in each chamber there being an ultrasonic standing wave that is moved transversely to its wave fronts, whereby said wave fronts are progressively displaced towards a chosen end region of the chamber, the conditions in the respective chambers being controlled to cause a selected type of particle in the fluid to become attached to the standing wave and be displaced towards said end region of each chamber and be concentrated at and/or collected from said end region, the successive chambers thereby separating the different selected particle types in the fluid.

2. A method according to claim 1 wherein in at least one of the chambers the selected particles are inspected and/or analysed while retained in said end region of the chamber by the standing wave therein.

3. A method according to claim 1 or claim 2 wherein the density of grouping of the selected particles is increased in said end region by the use of a plurality of non-coaxial standing waves and/or a convergent standing wave.

4. A method according to any one of the preceding claims wherein different ultrasonic wave systems are maintained in the respective chambers.

5. A method according to any one of the preceding claims wherein a first of the chambers is employed to concentrate particulate matter of a size substantially greater than the particle size in the or each succeeding chamber.

6. A method according to any one of the preceding claims wherein fluid from the or each preceding chamber to a succeeding chamber, and a fluid outlet from the final chamber, is taken from a position spaced from said end region of the preceding chamber.

7. A method according to any one of the preceding claims wherein at least one of the chambers has the standing wave established within

it with the axis of the wave extending in the direction of a maximum dimension of the chamber.

8. A method according to any one of claims 1 to 6 wherein the fluid flows through at least one of the chambers transversely to the standing wave.

9. A method according to claim 8 wherein the standing wave displaces the particles attached to it away from a direct path for said fluid flow between an inlet and outlet of said chamber.

10. A method according to claim 9 wherein different temperature regimes are established in said direct path and said end region whereby to inhibit diffusion of the fluid of said flow towards said end region.

11. A method according to any one of the preceding claims wherein selected particles in a body fluid are separated.

12. A method according to any one of the preceding claims for analysis of blood or urine.

13. Apparatus for separating a plurality of particle types present in a fluid flow, comprising a plurality of chambers arranged to receive a quantity of fluid in sequence, means for establishing in each chamber an ultrasonic standing wave and for moving said standing wave transversely to its wave fronts, whereby said wave fronts can be progressively displaced towards a chosen end region of the chamber, means for controlling the conditions in the respective chambers to cause selected particle types in the fluid to become attached to the standing wave so that the different selected particle types in the fluid can be concentrated at and/or collected from said end regions of the respective chambers.

14. Apparatus according to claim 13 comprising outlet means in said end region of at least one of the chambers for collecting the concentrated particles therefrom.

15. Apparatus according to claim 13 or claim 14 comprising means for inspecting and/or analysing the particles in said end region of at least one of the chambers.

16. Apparatus according to any one of claims 13 to 15 wherein generating means for said ultrasonic standing wave are arranged to operate at a frequency of at least 500kHz.

17. Apparatus according to any one of claims 13 to 16 comprising respective ultrasonic generating means for establishing different standing wave systems in the respective chambers.

18. Apparatus according to any one of claims 13 to 17 having means for producing non-coaxial standing waves and/or a convergent standing wave in at least one said end region for further concentration of the selected particles.

19. Apparatus according to any one of claims 13 to 18 wherein each chamber has an outlet for the fluid spaced from said end region of the chamber.

20. Apparatus according to any one of claims 13 to 19 wherein at least one of the chambers is elongate in the direction of propagation of the standing wav within it.

21. Apparatus according to any one of claims 13 to 19 wherein said means for establishing a standing wave in at least one chamber are arranged to establish the wave with its axis transverse to a fluid flow direction defined by inlet and outlet ports at opposite sides of the chamber.

22. Apparatus according to claim 21 wherein said ports are in an end region of the chamber remote from said end region in which the selected particle type is collected.

23. Apparatus according to claim 22 comprising means for maintaining a temperature difference between said end regions of said chamber to inhibit mixing of a fluid flow between said ports with the concentrated particles.

24. A methof of separating a plurality of particle types substantially as described herein.

25. Apparatus for separating a plurality of particle types constructed and arranged for use and operation substantially as described herein with reference to the accompanying drawings.

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